in the instrument. The view in FIG. **4** is also referred to as a sectional-isometric view of the cartridge lying over the heater wafer. A window **903** above the PCR channel in the cartridge is shown in perspective view. PCR channel **901** (for example, 150μ deep×700μ wide), is shown in an upper layer of the cartridge. A laminate layer **905** of the cartridge (for example, 125μ thick) is directly under the PCR channel **901**. As depicted, an optional further layer of thermal interface laminate **907** on the cartridge (for example, 125μ thick) lies directly under the laminate layer **905**. Heaters **909**, **911** are situated in a heater substrate layer **913** directly under the thermal interface laminate. The heaters are photolithographically defined and etched metal layers of gold (typically about 3,000 Å thick). Layers of 400 Å of TiW are deposited on top and bottom of the gold layer to serve as an adhesion layer. The substrate used is glass, fused silica or quartz wafer having a thickness of 0.4 mm, 0.5 mm, 0.7 mm, or 1 mm. A thin electrically-insulative layer of 2 μm silicon oxide serves as an insulative layer on top of the metal layer. Additional thin electrically insulative layers such as 2-4 g/m of Parylene may also be deposited on top of the silicon oxide surface. Two long heaters **909** and **911**, as further described herein, run alongside the PCR channel.

[0065] Referring to FIGS. **5**A and **5**B, an exemplary set of heaters configured to heat, cyclically, PCR reaction chamber **1001** is shown. It is to be understood that heater configurations to actuate other regions of a microfluidic cartridge such as other gates, valves, and actuators, may be designed and deployed according to similar principles to those governing the heaters shown in FIGS. **5**A and **5**B.

[0066] An exemplary PCR reaction chamber **1001** in a microfluidic substrate, typically a chamber or channel having a volume ~1.6 μl, is configured with a long side and a short side, each with an associated heating element. A PCR reaction chamber may also be referred to as a PCR reactor, herein, and the region of a cartridge in which the reaction chamber is situated may be called a zone. The heater substrate therefore preferably includes four heaters disposed along the sides of, and configured to heat, a given PCR reaction chamber, as shown in the exemplary embodiment of FIG. **5**A: long top heater **1005**, long bottom heater **1003**, short left heater **1007**, and short right heater **1009**. The small gap between long top heater **1005** and long bottom heater **1003** results in a negligible temperature gradient (less than 1° C. difference across the width of the PCR channel at any point along the length of the PCR reaction chamber) and therefore an effectively uniform temperature throughout the PCR reaction chamber. The heaters on the short edges of the PCR reactor provide heat to counteract the gradient created by the two long heaters from the center of the reactor to the edge of the reactor.

[0067] It would be understood by one of ordinary skill in the art that still other configurations of one or more heater(s) situated about a PCR reaction chamber are consistent with the methods and apparatus described herein. For example, a 'long' side of the reaction zone can be configured to be heated by two or more heaters. Specific orientations and configurations of heaters are used to create uniform zones of heating even on substrates having poor thermal conductivity because the poor thermal conductivity of glass, or quartz, polyimide, FR4, ceramic, or fused silica substrates is utilized to help in the independent operation of various microfluidic components such as valves and independent operation of the various PCR lanes. It would be further understood by one of ordinary skill in the art, that the principles underlying the configuration of heaters around a PCR reaction zone are similarly applicable to the arrangement of heaters adjacent to other components of the microfluidic cartridge, such as actuators, valves, and gates.

[0068] In certain embodiments, each heater has an associated temperature sensor. In the embodiment of FIG. **5**A, a single temperature sensor **1011** is used for both long heaters. A temperature sensor **1013** for short left heater, and a temperature sensor **1015** for short right heater are also shown. The temperature sensor in the middle of the reactor is used to provide feedback and control the amount of power supplied to the two long heaters, whereas each of the short heaters has a dedicated temperature sensor placed adjacent to it in order to control it. As further described herein, temperature sensors are preferably configured to transmit information about temperature in their vicinity to a processor in the apparatus at such times as the heaters are not receiving current that causes them to heat. This can be achieved with appropriate control of current cycles.

[0069] In order to reduce the number of sensor or heater elements required to control a PCR heater, the heaters may be used to sense as well as heat, and thereby obviate the need to have a separate dedicated sensor for each heater. In another embodiment, each of the four heaters may be designed to have an appropriate wattage, and connect the four heaters in series or in parallel to reduce the number of electronically-controllable elements from four to just one, thereby reducing the burden on the associated electronic circuitry.

[0070] FIG. **5**B shows expanded views of heaters and temperature sensors used in conjunction with a PCR reaction chamber of FIG. **5**A. Temperature sensors **1001** and **1013** are designed to have a room temperature resistance of approximately 200-300 ohms. This value of resistance is determined by controlling the thickness of the metal layer deposited (e.g., a sandwich of 400 Å TiW/3,000 Å Au/400 Å TiW), and etching the winding metal line to have a width of approximately 10-25 μm and 20-40 mm length. The use of metal in this layer gives it a temperature coefficient of resistivity of the order of 0.5-20° C./ohms, preferably in the range of 1.5-3° C./ohms. Measuring the resistance at higher temperatures enables determination of the exact temperature of the location of these sensors.

[0071] The configuration for uniform heating, shown in FIG. **5**A for a single PCR reaction chamber, can also be applied to a multi-lane PCR cartridge in which multiple independent PCR reactions occur.

[0072] Each heater can be independently controlled by a processor and/or control circuitry used in conjunction with the apparatus described herein. FIG. **5**C shows thermal images, from the top surface of a microfluidic cartridge when heated by heaters configured as in FIGS. **5**A and **5**B, when each heater in turn is activated, as follows: (A): Long Top only; (B) Long Bottom only; (C) Short Left only; (D) Short Right only; and (E) All Four Heaters on. Panel (F) shows a view of the reaction chamber and heaters on the same scale as the other image panels in FIG. **5**C. Also shown in the figure is a temperature bar.

Exemplary Microfluidic Cartridges

[0073] The multi-sample cartridge comprises at least a first microfluidic network and a second microfluidic network, adjacent to one another, wherein each of the first microfluidic network and the second microfluidic network is as elsewhere described herein, and wherein the first microfluidic network